# Effect of sulfate and iron(III) on LCFA degradation by a methanogenic community

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Under anaerobic conditions long chain fatty acids (LCFA) can be converted to methane by syntrophic bacteria and methanogenic archaea. LCFA degradation was also reported in the presence of alternative hydrogenotrophic partners, such as sulfate-reducing bacteria (SRB) and iron-reducing bacteria (IRB), which generally show higher affinity for H2 than methanogens and are more resistant to LCFA [1,2,3]. Their presence in a microbial culture degrading LCFA can be advantageous to reduce LCFA toxicity towards methanogens, although high concentrations of external electron acceptor (EEA) can lead to out-competition of methanogens and cease methane production. In this work, we tested the effect of adding sub-stoichiometric concentrations of sulfate and iron(III) to methanogenic communities degrading LCFA.

Methane production from oleate (3mM) was studied in batch vials with sulfate or ferric iron, and compared with methanogenic microcosms established in the absence of electron acceptors other than $CO_2$; the amount of EEA added is 1/3 of stoichiometric concentration needed for the complete oxidation of 3mM of oleate. Suspended sludge from a brewery wastewater treatment plant was used as inoculum. Reduction of electron acceptors, accumulation of fatty-acid intermediates, and production of $CH_4$ were monitored over time. Sulfate and iron were reduced during the first 25 days of incubation, probably due to H2 consumption. Acetate was concomitantly produced and accumulated in all the microcosms at concentrations close to stoichiometric values. This accumulation started earlier in the sulfate- and iron-reducing microcosms, suggesting a faster LCFA degradation than under methanogenic conditions. Further acetate consumption was controlled by acetoclastic methanogenesis, showing that SRB and IRB did not out-compete the methanogens. Methane production rate was not improved by the presence of the EEA, maybe due to inhibitory effects of $H_2S$ or Fe(III) towards acetoclastic methanogens.

These results highlight the complexity of anaerobic communities, in which metabolic pathways and microbial interactions are not thoroughly understood. Microbial composition in the different microcosms is under analysis by Illumina sequencing and may contribute for a better understanding of this complexity.

[1] Sousa *et al*.. Environ Microbiol 11(1), 2009, 68–80
[2] Coates *et al*.. Arch Microbiol, 164, 1995, 406–413
[3] Li *et al*. Water Res 39, 2005, 3109–3119